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Effect of urea fertilizer on the vitality of the earthworm Octolasion cyanieum

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A B S T R A C T

This study was conducted to find out the effect of rearing earthworms *O. cyanieum* in soil treated with concentrations (100, 250, 500 and 1000 mg / kg) for a period of (7, 14, 21 and 28 days) of rearing, as urea fertilizer had a significant effect on all activities The vital activity measured in the experiment, all worms died at a concentration of 1000 mg/kg after 7 days. There was also a decrease in weight, reaching the highest drop of 0.475 g, and the relative growth rate was 64.62% at a concentration of 500 mg/kg after 28 days. Also, the amount of protein content in the body of worms decreased with increasing concentrations.

Introduction

According to [2], [9], the United Nations stated that the world population is expected to increase to 9.8 billion in 2050, and the number will double in 2100 to reach 11.2 billion. Due to the increase in population, there is great pressure on agricultural land; Because they are so widely used to produce enough food, The agricultural soil must maintain sufficient levels of nutrients in quantity and quality for the production of food and energy. The use of mineral fertilizers has always been a major tool to compensate for the lack of nutrients for a plant and thus achieve an increase in yield. However, although chemical fertilizers are beneficial and are the main reason for producing sufficient crops For the world population, its overuse brings serious challenges for present and future generations such as air, water and soil pollution, degraded lands, depleted soils and increased greenhouse gas emissions. Not only do these synthetic fertilizers become dangerous to our environment, but also to humans, animals and other (beneficial) microbial life forms [6]. The long-term use of chemical fertilizers causes a decrease in soil pH, which leads to a deterioration in the physical, chemical and biological properties and crop quality, and thus harms soil organisms [8]. According to [12], chemical fertilizers are toxic to soil organisms such as earthworms, which are known to enhance soil fertility. [18] stated that earthworms are an important contributor to soil fertility in several ways, for example they raise nutrients. From the depths of the soil to the surface layer, earthworms mix soil layers, improve the distribution of organic matter in the soil, and make nutrients readily available to plants, as earthworms consume a lot of organic matter and soil and turn it into a nutrient-rich substance that is more water-soluble and readily available to plants.

The aim of this study was to find out the effect of earthworms *O cyanieum* rearing in the soil treated with urea fertilizer, by studying the weight rate, the relative growth rate, and the amount of protein content in the bodies of earthworms treated with fertilizer for a period of 28 days.

Materials and methods

Collecting earthworm samples:

This study was conducted at the University of Mosul, College of Education for Pure Sciences.Earthworms were collected in late August 2022 from home gardens in the Nimrud district south of Mosul / Nineveh, and more and were preserved and bred in plastic boxes with dimensions of (55 * 35 * 28 cm) (length * width * height). The boxes contained a Bird droppings were also placed with soil in these boxes for the purpose of feeding the worms during their rearing period according to the method [4; 3].

Fertilizers used in the study: in the current study, was used inorganic urea fertilizer, obtained from the local markets of the city of Mosul / Iraq, and the following selected concentrations (100, 250, 500 and 1000 mg / kg), The concentrations used in the study were estimated based on the recommended amounts per fertilizer kilogram/ Dunam, as well as selecting concentrations higher than the recommended amounts, As well as, the necessary fertilizer ratio for one treatment was determined by calculating the amount of soil in kilograms at a depth of 15 centimeters per Dunam, which was estimated at 500,000 kilograms per Dunam. By performing the ratio and proportion operation for each selected concentration of fertilizers, the necessary amount of fertilizer in grams per kilogram of soil was determined.

Preparation of biological effects experiments on earthworms *O. cyanieum*: A completely random block design with three replications and five urea fertilizer treatments, including one control treatment and four other treatments, was used. Plastic containers with a capacity of one kilogram were taken and filled with 500 grams of soil that had been previously sterilized at 90 degrees Celsius in an electric oven for five hours. The purpose of sterilization was to ensure that the soil was free of worm eggs or other organisms that could interfere with the experiment's results, Before treating the earthworms with fertilizer, they were placed in cups containing distilled water for over two hours to clean their digestive tract of soil and expel their waste. The worms' weights were then measured using a sensitive electronic scale, with 5 worms per repeater, ensuring they were approximately the same size and age, They were then placed in plastic containers containing soil treated with fertilizers, which had been mixed well. The plastic containers were then covered with a piece of transparent cloth to prevent the worms from escaping while being monitored for (7, 14, 21, and 28 days).

The effect of urea fertilizer concentrations on the weights of earthworms: Adult worms (with a clitellum) were selected, which were similar in weight and age, and their weights were calculated every week using a sensitive electronic scale, and the difference in their weights was noted compared to the control group.

The effect of urea fertilizer on the relative growth rate of earthworms: After measuring the weights of the worms before and after treatment with the urea fertilizer to determine the extent of the effect of urea fertilizer on the growth rate of the worms, their growth rate was calculated according to the method of [15] using the following equation:

Relative growth rate (RGR) = weight after treatment ÷ weight before treatment × 100.

Estimation of protein content in the bodies of earthworms: The amount of protein in the bodies of earthworms treated with urea fertilizer was estimated using the method of [10] and was modified by [14].

Statistical Analysis: The results were statistically analyzed using the SAS program on a computer with a completely randomized design. A factorial C.R.D. experiment was conducted, and the differences between the means were selected using a multiple range test (Duncan's test) at a 5% probability level [1].

Results

Effect raised earthworms *O. cyanieum* in soil treated with different concentrations of urea fertilizer and for different periods of time on earthworm weight:

The results in Table (1) show that treating soil with concentrations of 100, 250, 500, and 1000 mg /kg of urea fertilizer for cultivation periods of 7, 14, 21, and 28 days had a significant effect on the weight rate of *O. cyanieum* earthworms, We find from the table that the different concentrations had a clear effect, as the average weight decreased significantly and reached the lowest weight of 0.595 and 0.474 g at the concentrations of 500 and 1000 mg / kg, respectively. Both treatments different from the weight rate of the control treatment, which was 0.804 g.

As Table shows, the weight rate was also affected by the duration of the earthworms' stay in the environment treated with urea fertilizer, as the average weight after 7 days was 0.662 g, then it increased in the period of 14 and 21 days to 0.699 and 0.686 g, respectively, and decreased after 28 days to reach 0.663 g, and there was a difference between them moral.

As for the effect of the interaction between concentration and time period, we notice from Table (1) that at the time period of 7 days, the weight decreased significantly from 0.725 g at a concentration of 100 mg/kg to reach 0.474 g at a concentration of 1000 mg/kg, all concentrations except The concentration of 100 mg/kg recorded a significant difference with the control group in which the weight was 0.756 mg/kg. As for the second time period of 14 days, there was also a significant difference between the concentrations, as all worms died at the concentration of 1000 mg/kg, so the maximum weight recorded in this period was 0.735. g was at a concentration of 100 mg/kg, and the weight of the control group for the same time period was 0.771 g. As for the third time period of 21 days, we notice from the table that the weight of the worms decreased significantly to 0.716, 0.611 and 0.597 g at the concentrations of 100, 250 and 500 mg/kg respectively, As mentioned previously, all worms died at a concentration of 1000 mg/kg and the weight of the control group At this time it is 0.821 g. As for the time period of 28 days, we also notice a significant decrease in weight to reach 0.742, 0.568 and 0.475 g at concentrations of 100, 250 and 500 mg / kg, respectively, and the weight of the control group was 0.870 g for the same time period.

Table 1. The average weight (g) of the earthworm *O. cyanieum* raised in soil treated with different concentrations of urea fertilizer for several time periods.

Concentration (mg/kg)	Exposure period (days)				
	7	14	21	28	Concentration effect rate
100	0.725c	0.735c	0.716c	* 0.742c	0.729b
250	0.693cd	0.649de	0.611ef	0.568f	0.630c
500	0.666de	0.642e	0.597ef	0.475g	0.595d
1000	0.474g	……	……	** ……	0.474e
control	0.756c	0.771bc	0.821ab	0.870a	0.804a
Rate (time period effect)	0.662c	0.699a	0.686b	0.663c	

*Numbers that carry different letters have different meanings according to the Duncan multiple range test at a significance level of 0.05%.
** (…) means the death of all worms during the recorded period.

Effect of different concentrations of urea fertilizer on the relative growth rate of the earthworm *O. cyanieum*:

The results in Table (2) show that the soil treatment with different concentrations (100, 250, 500 and 1000 mg/kg) of urea fertilizer for periods of time (7, 14, 21 and 28 days) had a significant effect on the relative growth rate of earthworm *O cyanieum*, so we notice from the table that the different concentrations had a significant effect, so the relative growth rate was 95.85% at the concentration of 100 mg/kg and 63.36% at the concentration of 1000 mg/kg, while the relative growth rate in the control group was 110.05%. There was a significant difference between the two treatments.

Table (2) also shows that the duration of the transaction affected the relative growth rate, to rise initially from 89.21% in the first period of 7 days to reach 21.94% in the second period of 14 days, then it decreased in the third period of 21 days to 92.76% and continued. Declining to 89.68% in the fourth period of 28 days, with no significant difference between it and the first period.

As for the effect of the interaction between the concentration and the time period, we notice that in the first period of 7 days, the relative growth rate decreased significantly to 95.26% at the concentration of 100 mg / kg, reaching 63.36% at the concentration of 1000 mg / kg, and the relative growth rate in the control group was 103.41%. As for the second period of 14 days and the third period of 21 days, the relative growth rate also decreased in them, so the maximum relative growth rate for the period of 14 days was 96.58% at the concentration of 100 mg / kg, while all worms died in the concentration of 1000 mg / kg, and the control group recorded a relative rate The growth rate reached 105.47% for the same period of time, the maximum relative growth rate for a period of 21 days was 94.08% at a concentration of 100 mg / kg also, and the relative growth rate for the control group in this period amounted to 112.31%, and we also notice that in the fourth period of 28 days, the relative growth rate decreased significantly From 97.5% at a concentration of 100 mg/kg to 64.62% at a concentration of 500 mg/kg, but at a concentration of 1000 mg/kg all worms died as we mentioned earlier, and the relative growth rate of the control group in this period was 119.01%.

Table 2. Relative growth rate (%) of earthworm *O. cyanieum* raised in soil treated with different concentrations of urea fertilizer for various periods of time.

Concentration (mg/kg)	Exposure period (days)				
	7	14	21	28	Concentration effect rate
100	95.26ce	96.58cf	94.08eh	* 97.5fh	95.85b
250	94.67fh	88.66fi	83.46hi	77.59jk	86.09c
500	89.39fi	86.16gi	81.22ij	64.62kl	82.34d
1000	63.36l	.....	.....	**	63.36e
control	103.41cd	105.47bc	112.31ab	119.01a	110.05a
Rate (time period effect)	89.21c	94.21a	92.76b	89.68c	

*Numbers that carry different letters have different meanings according to the Duncan multiple range test at a significance level of 0.05%.
** (…) means the death of all worms during the recorded period.

Effect of urea fertilizer on the amount of protein content in the body of the earthworm *O. cyanieum*:

The results in Table (3) show the treatment of the soil with concentrations (100, 250, 500 and 1000 mg/kg) of urea fertilizer and its effect on the protein content in the body of the earthworm *O. cyanieum* after 28 days (except for the concentration of 1000 mg/kg in which the protein was measured After seven days), there was a significant difference in the amount of protein, which was recorded as 0.153, 0.119, 0.115 and 0.052 mg/g at concentrations of 100, 250, 500 and 1000 mg/kg respectively, and all of them differed significantly from the protein content of the control group 0.250 mg/g.

Table 3. Protein content (mg g) in the body of earthworms *O. cyanieum* raised in soil treated with different concentrations of urea fertilizer and for different periods of time.

	control	100	250	500	1000
The amount of protein	0.250a	0.153b	0.119c	0.115c	0.052 *

*Numbers that carry different letters have different meanings according to the Duncan multiple range test at a significance level of 0.05%

Discussion

Effect of urea fertilizer on weight and relative growth rate of earthworm *O. cyanieum*:

From the results in Table (1) that the soil was treated with concentrations (100, 250, 500 and 1000 mg/kg) of urea fertilizer and for the cultivation time periods (7, 14, 21 and 28 days), we note that the weights of worms in all the concentrations mentioned above were It decreases as the concentration increases and the duration of exposure increases, as well as the relative growth rate of earthworms was affected in the results of Table (2) of soil treatment at concentrations (100, 250, 500 and 1000 mg/kg) with urea fertilizer and for the cultivation time periods (7, 14, 21 and 28). days), as the decrease in the relative growth rate of worms was increasing with increasing concentration and cultivation time period. It has been concluded that the addition of urea fertilizer to the soil leads to changes in soil chemistry, such as a change in soil pH and a difference in ammonia levels, and it was also found that these changes have an effect on soil organisms, including earthworms [16] Our study coincided with [11]. As the application of urea on soil containing earthworms reduced the weight of earthworms, and our study converged with the study of [17] as the average weight of earthworm decreased when the soil in which earthworms were reared was treated with polycyclic aromatic hydrocarbons (piperine). Our results are similar to that of [5], where exposure of earthworms *Eisenia. fetida* (14 days) to oxybenzoin inhibited growth rate.

Effect of urea fertilizer on the protein content in the body of earthworms

From the results in Table (3), it was shown that soil treatment with concentrations (100, 250, 500 and 1000 mg/kg) of urea fertilizer negatively affected the protein content in the body of the earthworm. The results of [13] are similar to our study in their study of the effect of pesticides. Glyphosate and Gonocrotophos in the bodies of earthworms *Drawida*. *willsi* and *Lampito mauritii*, as the protein content decreased in the bodies of worms *L. mauritii* with increasing concentrations of Glyphosate. The current study also agreed with the study of (7), as the amount of protein decreased after 28 days of treating earthworms with concentrations 2 and 10 mg/kg of atrazine herbicide compared with the control group.

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